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Michael L. Goldman
NIXON PEABODY LLP
Clinton Square
P.O. Box 31051
Rochester, NY 14603

EXAMINER
FREDMAN, JEFFREY NORMAN

ART UNIT	PAPER NUMBER
1634	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/033,300	ZHANG ET AL.	
	Examiner	Art Unit	
	Jeffrey Fredman	1634	

-- Th MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *28 July 2003* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-87* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-87* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☒ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

Election/Restrictions

1. The restriction requirement between Groups I and II is withdraw, since Applicant correctly notes that the claims are not independent and distinct.

Claim Rejections - 35 USC § 112

2. Claims 43, 65 and 87 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 43, 65 and 87 are vague and indefinite because it is unclear how the step of electrospraying can be performed in the same container as the enzymatic assay steps. In particular, the claims as written are open to at least two possible interpretations. The first is that when the claim requires that "said method is carrier out in a single container", the claim is only referring to the enzymatic steps and not the detection steps in which the solution is electrosprayed into a chromatography reagent. The second is that the claim are intended to mean some sort of integrated system. However, since the claims are not currently claiming such a system, and the word "container" would be difficult to interpret to mean such a system, the first interpretation will be used for the prior art rejections. Claim 22 is not included in this rejection because that claim can be interpreted as solely performing the claimed enzymatic steps in a single container, which is definite and which is anticipated as discussed below.

Claim Rejections - 35 USC § 102

3. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

4. Claims 1-3, 5-7, 9, 23-25, 27 and 30 are rejected under 35 U.S.C. 102(b) as being anticipated by Hoogendoorn et al (Human Genetics (1999) 104:89-93).

Hoogendoorn teaches a method of detecting single nucleotide polymorphisms (see abstract) comprising:

(a) providing a sample potentially containing a target nucleic acid molecule (see page 90, column 1, subheading "Target sequences and primers"),

(b) subjecting the sample to a polymerase chain reaction process involving use of oligonucleotide amplification primers under conditions effective to amplify any of the target nucleic acid molecule present in the sample to produce an amplification product (see page 90, columns 1 and 2, subheading "PCR chain reaction"),

(c) subjecting the amplification product to treatment with a phosphatase under conditions effective to remove 5' phosphates from free deoxynucleotide triphosphates (dNTPs) in the amplification product (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension")

(d) inactivating the phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension)

(e) providing an oligonucleotide extension primer complementary to a portion of the target nucleic acid molecule (see page 90, column 2, subheading "Primer extension reactions")

(f) providing a nucleic acid polymerizing enzyme (see page 90, column 2, subheading "Primer extension reactions")

(g) providing a plurality of types of nucleotide analogs (see page 90, column 2, subheading "Primer extension reactions")

(h) blending the amplification product treated with a phosphatase, the oligonucleotide extension primer, the nucleic acid polymerizing enzyme, and the nucleotide analogs, each type being present in a first amount, to form an extension solution where the oligonucleotide extension primer is hybridized to the target nucleic acid molecule to form a primed target nucleic acid molecule and the nucleic acid polymerizing enzyme is positioned to add nucleotide analogs to the primed target nucleic acid molecule at an active site (see page 90, column 2, subheading "Primer extension reactions")

(i) extending the oligonucleotide extension primer in the extension solution by using the nucleic acid polymerizing enzyme to add a nucleotide analog to the oligonucleotide extension primer at the active site to form an extended oligonucleotide extension primer, wherein the nucleotide analog being added is complementary to the nucleotide of the target nucleic acid molecule at the active site (see page 90, column 2, subheading "Primer extension reactions")

(j) determining the amounts of each type of the nucleotide analogs in the extension solution after said extending, each type being present in a second amount (see page 90, subheading "HPLC analysis" to page 91, figures 2 and 3)

(k) comparing the first and second amount of each type of the nucleotide analog (see page 91, figures 2 and 3)

(l) and identifying the type of nucleotide analog where the first and second amounts differ as the nucleotide added to the oligonucleotide extension primer at the active site (see page 91, figures 2 and 3).

With regard to claims 2-3, 24-25, Hoogendoorn teaches the use of genomic DNA, which will comprise both double and single stranded DNA (see page 90, column 1, subheading "PCR chain reaction").

With regard to claim 5, 27, Hoogendoorn teaches the use of primers with different Tms where the NT1-1a(L) primer of page 90 on table 1, used for amplification, has a basic Tm of 52 C, while the NT1-PEXT primer has a basic Tm of 48 C as calculated by http://www.basic.nwu.edu/biotools/oligocalc.html.

With regard to claim 6, Hoogendoorn teaches the use of Shrimp alkaline phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claim 7, Hoogendoorn teaches digesting the PCR product with exonuclease I to digest primers which do not produce an amplification product, followed by inactivation of the exonuclease I (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claim 9, 30, Hoogendoorn teaches the use of ddNTP analogs (see page 90, column 2, subheading "primer extension reactions")

With regard to claim 23, Hoogendoorn teaches the steps of claim 1 as discussed above, and Hoogendoorn further teaches replacing steps (c) and (d) which remove primers with the step of passing the amplification product through a DNA purification column which retains the amplified DNA but not the primers (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

5. Claims 23-25, 27 and 30 are rejected under 35 U.S.C. 102(b) as being anticipated by Higgins et al (Biotechniques (1997) 23(4):710-713)

Higgins teaches a method of detecting single nucleotide polymorphisms (see abstract) comprising:

(a) providing a sample potentially containing a target nucleic acid molecule (see page 710, column 2, subheading "PCR amplification and strand immobilization"),

(b) subjecting the sample to a polymerase chain reaction process involving use of oligonucleotide amplification primers under conditions effective to amplify any of the target nucleic acid molecule present in the sample to produce an amplification product (see page 710, column 2, subheading "PCR amplification and strand immobilization"),

(c) passing the amplification product through a molecular weight filter configured to retain amplified target nucleic acid but not the amplification primers (see page 710, column 2, subheading "PCR amplification and strand immobilization")

(d) providing an oligonucleotide extension primer complementary to a portion of the target nucleic acid molecule (see page 710, column 3, subheading "COSBE conditions"),

(e) providing a nucleic acid polymerizing enzyme (see page 710, column 3, subheading "COSBE conditions"),

(f) providing a plurality of types of nucleotide analogs (see page 710, column 3, subheading "COSBE conditions" which teaches the use of dGTP and page 711, column 1, which teaches the use of ddNTPs),

(g) blending the amplification product treated with a phosphatase, the oligonucleotide extension primer, the nucleic acid polymerizing enzyme, and the nucleotide analogs, each type being present in a first amount, to form an extension solution where the oligonucleotide extension primer is hybridized to the target nucleic acid molecule to form a primed target nucleic acid molecule and the nucleic acid polymerizing enzyme is positioned to add nucleotide analogs to the primed target nucleic acid molecule at an active site (see page 710, column 3, subheading "COSBE conditions"),

(h) extending the oligonucleotide extension primer in the extension solution by using the nucleic acid polymerizing enzyme to add a nucleotide analog to the oligonucleotide extension primer at the active site to form an extended oligonucleotide extension primer, wherein the nucleotide analog being added is complementary to the nucleotide of the target nucleic acid molecule at the active site (see page 710, column 3, subheading "COSBE conditions"),

(j) determining the amounts of each type of the nucleotide analogs in the extension solution after said extending, each type being present in a second amount (see figure 2)

(k) comparing the first and second amount of each type of the nucleotide analog (see figure 2)

(l) and identifying the type of nucleotide analog where the first and second amounts differ as the nucleotide added to the oligonucleotide extension primer at the active site (see figure 2).

With regard to claims 24-25, Higgins teaches the use of genomic DNA, which will comprise both double and single stranded DNA (see page 710, column 1).

With regard to claim 27, Higgins teaches the use of primers with different Tms where the Ex10 PCR primer of Table I, used for amplification, has a basic Tm of 54 C, while the COSBE F508 M primer has a basic Tm of 55 C as calculated by http://www.basic.nwu.edu/biotools/oligocalc.html.

With regard to claim 30, Higgins teaches the use of ddNTP analogs (see page 711, column 1)

6. Claims 1-3, 5-7, 9 and 22 are rejected under 35 U.S.C. 102(b) as being anticipated by Chen et al (Genome Research (1999) 9:492-498).

Chen teaches a method of detecting single nucleotide polymorphisms (see abstract) comprising:

(a) providing a sample potentially containing a target nucleic acid molecule (see page 497, column 2, subheading "PCR amplification"),

(b) subjecting the sample to a polymerase chain reaction process involving use of oligonucleotide amplification primers under conditions effective to amplify any of the target nucleic acid molecule present in the sample to produce an amplification product (see page 497, column 2, subheading "PCR amplification"),

(c) subjecting the amplification product to treatment with a phosphatase under conditions effective to remove 5' phosphates from free deoxynucleotide triphosphates (dNTPs) in the amplification product (see page 497, column 2, subheading "Primer and dNTP degradation")

(d) inactivating the phosphatase (see page 497, column 2, subheading "Primer and dNTP degradation")

(e) providing an oligonucleotide extension primer complementary to a portion of the target nucleic acid molecule (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1)

(f) providing a nucleic acid polymerizing enzyme (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1),

(g) providing a plurality of types of nucleotide analogs (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1),

(h) blending the amplification product treated with a phosphatase, the oligonucleotide extension primer, the nucleic acid polymerizing enzyme, and the nucleotide analogs, each type being present in a first amount, to form an extension solution where the oligonucleotide extension primer is hybridized to the target nucleic acid molecule to form a primed target nucleic acid molecule and the nucleic acid polymerizing enzyme is positioned to add nucleotide analogs to the primed target nucleic acid molecule at an active site (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1)

(i) extending the oligonucleotide extension primer in the extension solution by using the nucleic acid polymerizing enzyme to add a nucleotide analog to the oligonucleotide extension primer at the active site to form an extended oligonucleotide extension primer, wherein the nucleotide analog being added is complementary to the nucleotide of the target nucleic acid molecule at the active site (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1)

(j) determining the amounts of each type of the nucleotide analogs in the extension solution after said extending, each type being present in a second amount (see page 498, column 1, subheading "Fluorescence polarization measurement" and figure 4)

(k) comparing the first and second amount of each type of the nucleotide analog (see figure 4)

(I) and identifying the type of nucleotide analog where the first and second amounts differ as the nucleotide added to the oligonucleotide extension primer at the active site (see figure 4).

With regard to claims 2-3, Chen teaches the use of genomic DNA, which will comprise both double and single stranded DNA (see page 494, column 1 and page 497, column 2, subheading "PCR amplification").

With regard to claim 5, Chen teaches the use of primers with different Tms where the C282Y-P1 primer of Table 1, used for amplification, has a basic Tm of 52 C, while the C282Y-31 primer has a basic Tm of 66 C as calculated by http://www.basic.nwu.edu/biotools/oligocalc.html.

With regard to claim 6, Chen teaches the use of Shrimp alkaline phosphatase (see page 497, column 2, subheading "Primer and dNTP degradation").

With regard to claim 7, Chen teaches digesting the PCR product with exonuclease I to digest primers which do not produce an amplification product, followed by inactivation of the exonuclease I ((see page 497, column 2, subheading "Primer and dNTP degradation").

With regard to claim 9, Chen teaches the use of ddNTP analogs (see page 497, column 2, subheading "Genotyping by the FP-TDI assay" to page 498, column 1),

With regard to claim 22, Chen teaches performing the assay in a single container (see abstract).

Claim Rejections - 35 USC § 103

7. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

8. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

9. Claims 1-9, 23-28 and 30 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hoogendoorn et al (Human Genetics (1999) 104:89-93) in view of Higuchi et al (Nucleic Acids Research (1989) 17:5865).

Hoogendoorn teaches a method of detecting single nucleotide polymorphisms (see abstract) comprising:

(a) providing a sample potentially containing a target nucleic acid molecule (see page 90, column 1, subheading "Target sequences and primers"),

(b) subjecting the sample to a polymerase chain reaction process involving use of oligonucleotide amplification primers under conditions effective to amplify any of the

target nucleic acid molecule present in the sample to produce an amplification product (see page 90, columns 1 and 2, subheading "PCR chain reaction"),

(c) subjecting the amplification product to treatment with a phosphatase under conditions effective to remove 5' phosphates from free deoxynucleotide triphosphates (dNTPs) in the amplification product (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension")

(d) inactivating the phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension)

(e) providing an oligonucleotide extension primer complementary to a portion of the target nucleic acid molecule (see page 90, column 2, subheading "Primer extension reactions")

(f) providing a nucleic acid polymerizing enzyme (see page 90, column 2, subheading "Primer extension reactions")

(g) providing a plurality of types of nucleotide analogs (see page 90, column 2, subheading "Primer extension reactions")

(h) blending the amplification product treated with a phosphatase, the oligonucleotide extension primer, the nucleic acid polymerizing enzyme, and the nucleotide analogs, each type being present in a first amount, to form an extension solution where the oligonucleotide extension primer is hybridized to the target nucleic acid molecule to form a primed target nucleic acid molecule and the nucleic acid polymerizing enzyme is positioned to add nucleotide analogs to the primed target

nucleic acid molecule at an active site (see page 90, column 2, subheading "Primer extension reactions")

(i) extending the oligonucleotide extension primer in the extension solution by using the nucleic acid polymerizing enzyme to add a nucleotide analog to the oligonucleotide extension primer at the active site to form an extended oligonucleotide extension primer, wherein the nucleotide analog being added is complementary to the nucleotide of the target nucleic acid molecule at the active site (see page 90, column 2, subheading "Primer extension reactions")

(j) determining the amounts of each type of the nucleotide analogs in the extension solution after said extending, each type being present in a second amount (see page 90, subheading "HPLC analysis" to page 91, figures 2 and 3)

(k) comparing the first and second amount of each type of the nucleotide analog (see page 91, figures 2 and 3)

(l) and identifying the type of nucleotide analog where the first and second amounts differ as the nucleotide added to the oligonucleotide extension primer at the active site (see page 91, figures 2 and 3).

With regard to claims 2-3, 24-25, Hoogendoorn teaches the use of genomic DNA, which will comprise both double and single stranded DNA (see page 90, column 1, subheading "PCR chain reaction").

With regard to claim 5, 27, Hoogendoorn teaches the use of primers with different Tms where the NT1-1a(L) primer of page 90 on table 1, used for amplification,

has a basic Tm of 52 C, while the NT1-PEXT primer has a basic Tm of 48 C as calculated by http://www.basic.nwu.edu/biotools/oligocalc.html.

With regard to claim 6, Hoogendoorn teaches the use of Shrimp alkaline phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claim 7, Hoogendoorn teaches digesting the PCR product with exonuclease I to digest primers which do not produce an amplification product, followed by inactivation of the exonuclease I (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claim 9, 30, Hoogendoorn teaches the use of ddNTP analogs (see page 90, column 2, subheading "primer extension reactions")

With regard to claim 23, Hoogendoorn teaches the steps of claim 1 as discussed above, and Hoogendoorn further teaches replacing steps (c) and (d) which remove primers with the step of passing the amplification product through a DNA purification column which retains the amplified DNA but not the primers (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

Hoogendoorn does not teach application of the method to RNA nor does Hoogendoorn teach the use of lambda exonuclease to generate single strands.

Higuchi teaches formation of single strands by lambda exonuclease digestion (see page 5865).

It would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to study RNA by the method of Hoogendoorn where

the SNP of interest would be found in the RNA sequence since an ordinary practitioner would have recognized that RNA is an equivalent nucleic acid template to DNA for detection of SNPs. Further, it would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to modify the method of Hoogendoorn to use the single strands of Higuchi since Higuchi notes that the method assists in sequence analysis (see page 5865 "Therefore, the strand of DNA synthesized from the phosphorylated primer is digested by the exonuclease, leaving the complementary strand which can be sequenced by standard methods) such as the single nucleotide primer extension of Hoogendoorn. An ordinary practitioner would have been motivated to combine these methods to obtain single strands from the PCR reaction of Hoogendoorn to improve the efficiency of the primer extension assay of Hoogendoorn, by providing single stranded DNA template for detection.

10. Claims 1-7, 9-21, 23-28, 30-50, 52-63, 66-70 and 73-85 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hoogendoorn et al (Human Genetics (1999) 104:89-93) in view of Moon et al (U.S. Patent 6,569,324).

Hoogendoorn teaches a method of detecting single nucleotide polymorphisms (see abstract) comprising:

(a) providing a sample potentially containing a target nucleic acid molecule (see page 90, column 1, subheading "Target sequences and primers"),

(b) subjecting the sample to a polymerase chain reaction process involving use of oligonucleotide amplification primers under conditions effective to amplify any of the

target nucleic acid molecule present in the sample to produce an amplification product (see page 90, columns 1 and 2, subheading "PCR chain reaction"),

(c) subjecting the amplification product to treatment with a phosphatase under conditions effective to remove 5' phosphates from free deoxynucleotide triphosphates (dNTPs) in the amplification product (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension")

(d) inactivating the phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension)

(e) providing an oligonucleotide extension primer complementary to a portion of the target nucleic acid molecule (see page 90, column 2, subheading "Primer extension reactions")

(f) providing a nucleic acid polymerizing enzyme (see page 90, column 2, subheading "Primer extension reactions")

(g) providing a plurality of types of nucleotide analogs (see page 90, column 2, subheading "Primer extension reactions")

(h) blending the amplification product treated with a phosphatase, the oligonucleotide extension primer, the nucleic acid polymerizing enzyme, and the nucleotide analogs, each type being present in a first amount, to form an extension solution where the oligonucleotide extension primer is hybridized to the target nucleic acid molecule to form a primed target nucleic acid molecule and the nucleic acid polymerizing enzyme is positioned to add nucleotide analogs to the primed target

nucleic acid molecule at an active site (see page 90, column 2, subheading "Primer extension reactions")

(i) extending the oligonucleotide extension primer in the extension solution by using the nucleic acid polymerizing enzyme to add a nucleotide analog to the oligonucleotide extension primer at the active site to form an extended oligonucleotide extension primer, wherein the nucleotide analog being added is complementary to the nucleotide of the target nucleic acid molecule at the active site (see page 90, column 2, subheading "Primer extension reactions")

(j) determining the amounts of each type of the nucleotide analogs in the extension solution after said extending, each type being present in a second amount (see page 90, subheading "HPLC analysis" to page 91, figures 2 and 3)

(k) comparing the first and second amount of each type of the nucleotide analog (see page 91, figures 2 and 3)

(l) and identifying the type of nucleotide analog where the first and second amounts differ as the nucleotide added to the oligonucleotide extension primer at the active site (see page 91, figures 2 and 3).

With regard to claims 2-3, 24-25, 45-46, 67-68, Hoogendoorn teaches the use of genomic DNA, which will comprise both double and single stranded DNA (see page 90, column 1, subheading "PCR chain reaction").

With regard to claims 5, 27, 48, 70, Hoogendoorn teaches the use of primers with different Tms where the NT1-1a(L) primer of page 90 on table 1, used for amplification,

has a basic Tm of 52 C, while the NT1-PEXT primer has a basic Tm of 48 C as calculated by http://www.basic.nwu.edu/biotools/oligocalc.html.

With regard to claims 6, 49, Hoogendoorn teaches the use of Shrimp alkaline phosphatase (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claims 7, 50, Hoogendoorn teaches digesting the PCR product with exonuclease I to digest primers which do not produce an amplification product, followed by inactivation of the exonuclease I (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claims 9, 30, 52, Hoogendoorn teaches the use of ddNTP analogs (see page 90, column 2, subheading "primer extension reactions")

With regard to claims 23, 63, 85, Hoogendoorn teaches the steps of claim 1 as discussed above, and Hoogendoorn further teaches replacing steps (c) and (d) which remove primers with the step of passing the amplification product through a DNA purification column which retains the amplified DNA but not the primers (see page 90, column 2, subheading "Preparation of PCR fragments for primer extension").

With regard to claims 19, 20, 40, 41, 61, 62, 73, 82, 83, Hoogendoorn teaches detection of the nucleotide analogs in the liquid chromatography step (see page 91, figures 2 and 3).

With regard to claim 21, 42, 84, Hoogendoorn teaches the use of a DNAsep column which comprises a guard cartridge that comprises a chelating resin (see page 90, column 2).

Hoogendoorn does not teach using an electrospray device for detection of the primer extension products in association with the liquid chromatography detection.

Moon teaches, with regard to claims 10, 11, 31, 32, 53, 74, the use of an electrospray device with liquid chromatography (see abstract), wherein the electrospray device comprises:

(a) a substrate having an injection surface and an ejection surface opposite the injection surface (see column 11, lines 15-37) wherein the substrate is an integral monolith (see abstract) further comprising:

(i) an entrance orifice on the injection surface (see column 11, line 4 and figures 2-4),

(ii) an exit orifice on the ejection surface (see column 11, line 5 and figures 2-4),

(iii) a channel extending between the entrance orifice and the exit orifice (see column 11, lines 3-4 and figures 2-4),

(iv) a recess extending into the ejection surface and surrounding the exit orifice, thereby defining a nozzle on the ejection surface (see column 11, lines 5-48 and figures 2-4).

With regard to claim 12, 33, 54, 75, Moon further teaches a plurality of entrance and exit orifices connected by a plurality of channels (see figure 23 and column 22, lines 5-33).

With regard to claim 13, 34, 55, 76, Moon further teaches a voltage application system comprising a first and second electrode to define an electric field surrounding the exit orifice (see column 12, lines 1-67 and figures 2-4).

With regard to claim 14, 35, 56, 77, Moon teaches isolation of the electrode (see column 12, lines 40-50 and figures 2-4).

With regard to claim 15-16, 36-37, 57-58, 78-79, Moon teaches the situation where the electrodes are in contact with the solution (see column 12, lines 1-6 and figures 2-5).

With regard to claim 17, 38, 59, 80, Moon teaches modifying the electrodes to achieve the size of the fluid flow desired (see column 13, lines 5-45).

With regard to claim 18, 39, 60, 81, Moon teaches combining the electrospray device with a liquid chromatography device (see column 23, line 8 to column 24, line 67, for example).

With regard to claims 44 and 66, Hoogendoorn teaches the steps of claims 1-3, 5-7, 9, 23-25, 27 and 30 as discussed above and Moon teaches electrospraying as discussed above.

It would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to study RNA by the method of Hoogendoorn where the SNP of interest would be found in the RNA sequence since an ordinary practitioner would have recognized that RNA is an equivalent nucleic acid template to DNA for detection of SNPs.

Further, it would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to use the electrospray apparatus and method of Moon to prepare the liquid chromatography sample of Hoogendoorn for injection of the primer extension products of Hoogendoorn in the liquid chromatography column since Moon states "It also would be desirable to provide rapid sequential analysis of compounds which interact with a gene (see column 2, lines 51-52)" and Moon later notes regarding the electrospray apparatus that "The microchip based electrospray ionization device of the present invention provides minimal extra column dispersion as a result of a reduction in the extra column volume and provides efficient, reproducible and reliable and rugged formation of an electrospray. (see column 7, lines 43-46)." An ordinary practitioner would have been motivated to make the method of liquid chromatography method of Hoogendoorn more efficient and less costly by combining it with the device of Moon in which a integral monolithic electrospray device is mated to a liquid chromatography column to achieve efficient, reproducible and reliable results with reduced column volume, thereby improving efficacy while reducing cost.

Claims 1-21, 23-28, 30-63, 66-71 and 73-85 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hoogendoorn et al (Human Genetics (1999) 104:89-93) in view of Moon et al (U.S. Patent 6,569,324) and further in view of Higuchi.

Hoogendoorn in view of Moon teach the limitations of claims 1-7, 9-21, 23-28, 30-50, 52-63, 66-70 and 73-85 as discussed above. Hoogendoorn in view of Moon do not teach the use of lambda exonuclease to generate single strands.

Higuchi teaches formation of single strands by lambda exonuclease digestion (see page 5865).

It would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to modify the method of Hoogendoorn in view of Moon to use the single strands of Higuchi since Higuchi notes that the method assists in sequence analysis (see page 5865 "Therefore, the strand of DNA synthesized from the phosphorylated primer is digested by the exonuclease, leaving the complementary strand which can be sequenced by standard methods) such as the single nucleotide primer extension of Hoogendoorn. An ordinary practitioner would have been motivated to combine these methods to obtain single strands from the PCR reaction of Hoogendoorn to improve the efficiency of the primer extension assay of Hoogendoorn in view of Moon, by providing single stranded DNA template for detection.

11. Claims 1-7, 9-21, 23-50, 52-64, 66-70 and 72-86 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hoogendoorn et al (Human Genetics (1999) 104:89-93) in view of Moon et al (U.S. Patent 6,569,324) and further in view of Padhye et al (U.S. Patent 5,658,548).

Hoogendoorn in view of Moon teach the limitations of claims 1-7, 9-21, 23-28, 30-50, 52-63, 66-70 and 73-85 as discussed above. Hoogendoorn in view of Moon do not teach the use of a vacuum manifold for separation.

Padhye teaches the use of a vacuum manifold for use with the DNA purification columns of Hoogendoorn for the separation of PCR products from primers (see column 17, example 7, and column 7, line 59, in particular).

It would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to modify the method of Hoogendoorn in view of Moon to use the vacuum manifold of Padhye in the column purification method of Hoogendoorn since Padhye states that the use of the vacuum manifold will permit separation of the purified product from the impure protein and other contaminants (see column 8). An ordinary practitioner would have been motivated to use the vacuum manifold of Padhye with the column of Hoogendoorn in view of Moon in order to rapidly add the liquid through the column and rapidly separate the materials within the column.

12. Claims 1-7, 9-28, 30-50, 52-63, 65-70, 73-85 and 87 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hoogendoorn et al (Human Genetics (1999) 104:89-93) in view of Moon et al (U.S. Patent 6,569,324) and further in view of Chen et al (Genome Research (1999) 9:492-498).

Hoogendoorn in view of Moon teach the limitations of claims 1-7, 9-21, 23-28, 30-50, 52-63, 66-70 and 73-85 as discussed above. Hoogendoorn in view of Moon do not teach performance of the method in a single container. While some of these claims are unclear as to what is meant by a "single container", the rejection will assume that the enzymatic reaction proceeds in a single container.

Chen teaches performing the assay in a single container (see abstract)

It would have been prima facie obvious to one having ordinary skill in the art at the time the invention was made to modify the method of Hoogendoorn in view of Moon to perform the method in a single container since Chen expressly states that the mixture can be "analyzed directly without separation or purification" (see abstract). An ordinary practitioner would have been motivated to perform the method in a single container in order to minimize the need for separation and purification as taught by Chen, as well as Chen expressly notes that this method will reduce the amount of laboratory skill and minimize human handling necessary, which will reduce the possibility of contamination of the sample or other errors.

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Jeffrey Fredman whose telephone number is 703-308-6568. The examiner can normally be reached on 6:30-4:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 703-308-1119. The fax phone number for the organization where this application or proceeding is assigned is (703) 872-9306.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-308-0196.

Jeffrey Fredman
Primary Examiner
Art Unit 1634